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In Vitro Activities of Trovafloxacin against 557 Strains of Anaerobic Bacteria

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The antimicrobial activity of trovafloxacin for 557 strains of anaerobic bacteria was determined by the National Committee for Clinical Laboratory Standards-approved Wadsworth agar dilution technique. The species tested included *Bacteroides fragilis* ($n = 91$), other members of the *B. fragilis* group ($n = 130$), *Campylobacter gracilis* ($n = 15$), other *Bacteroides* spp. ($n = 16$), *Prevotella* spp. ($n = 49$), *Porphyromonas* spp. ($n = 15$), *Fusobacterium* spp. ($n = 62$), *Bilophila wadsworthia* ($n = 24$), *Sutterella wadsworthensis* ($n = 21$), *Clostridium* spp. ($n = 61$), *Peptostreptococcus* spp. ($n = 38$), and gram-positive non-spore-forming rods ($n = 35$). Trovafloxacin inhibited all strains of *B. fragilis* at ≤0.5 μg/ml, 99% of other *B. fragilis* group species at ≤2 μg/ml, and 96% of all anaerobes tested at ≤2 μg/ml.

Bacteroides fragilis group organisms have been tested with a variety of quinolone agents, including lomefloxacin, ciprofloxacin, norfloxacin, pefloxacin, nalidixic acid, enoxacin, cinoxacin, difloxacin, and ofloxacin, and are generally considered resistant to these agents (3, 4, 7, 9, 10, 16, 19). Although one study reported that all isolates of *B. fragilis* tested were susceptible to 16 mg of ciprofloxacin and ofloxacin per liter and 68 and 81% of isolates were susceptible to less than 4 mg/liter, respectively (5), the current approved aerobic breakpoint for most of these agents is 2 μg/ml. Activity against *Prevotella* species, such as the *Prevotella melaninogenicus-P. oralis* group, and against the *Bacteroides ureolyticus* group is generally more variable (9). We have recently reported the anti-anaerobic-bacterium activities of sparfloxacin and WIN 57273 (20). Sparfloxacin at 2 μg/ml inhibited 78% of *B. fragilis* strains and 44% of other *B. fragilis* group isolates. WIN 57273 displayed excellent activity against anaerobes, inhibiting all strains of the *B. fragilis* group at 2 μg/ml. Only two strains of *Fusobacterium* species were resistant (MICs, 4 μg/ml). Other investigators have found similarly good activity with WIN 57273 (8, 18). Clinafloxacin has shown excellent activity against anaerobes in a number of studies (1, 2, 6, 14), including our own (21). The Daiichi compound DU 67859a also inhibited nearly all anaerobes tested by us (22) and investigators at other institutions (11, 13, 17).

All bacteria were randomly selected recent clinical isolates from the Veterans Administration Wadsworth Medical Center, Los Angeles, Calif. The bacteria were identified according to established procedures (8, 15). The MICs were determined by an agar dilution technique described previously (15) with an inoculum of 10^5 CFU and brucella base-laked blood agar. The plates were incubated in an anaerobic chamber (Anaerobe Systems, San Jose, Calif.) for 48 h at 37°C. MICs were defined as the lowest concentration of antimicrobial agent resulting in no growth, a haze, one discrete colony or multiple tiny colonies, or a marked change in the appearance of growth compared with the appearance of the growth on the control plate (in the case of persistent light [slight] growth) (12). Reference strains of *B. fragilis* (ATCC 25285) and *Bacteroides thetaiotaomicron* (ATCC 29741) were used as controls in each test. The following antimicrobial agents were obtained as powders from the indicated companies: trovafloxacin, Pfizer Pharmaceuticals, New York, N.Y.; ciprofloxacin, Miles Pharmaceuticals, West Haven, Conn.; imipenem, Merck Sharp & Dohme, Rahway, N.J.; and metronidazole, Sigma, St. Louis, Mo.

The results of the studies are presented in Table 1. The percentages of organisms susceptibile at concentration ranges corresponding to three twofold dilutions are indicated in Table 1. The range for each antimicrobial agent is listed in footnote *b* of Table 1. The data are reported in this manner to mitigate the interpretive problems caused by the degree of acceptable variation in a twofold dilution system combined with the clustering of MICs near the breakpoint for many antimicrobial agent-anaerobe combinations. Since no breakpoint for anaerobic bacteria has been approved for quinolones, the data are not interpreted in categorical terms. If enough strains of one species were tested to give meaningful results, those data are listed separately.

Trovafloxacin inhibited all of the strains of *B. fragilis* tested at 0.5 μg/ml and 96% of the other *B. fragilis* group species tested at ≤2 μg/ml. For two strains of *Bacteroides distasonis* and two strains of *B. thetaiotaomicron*, trovafloxacin MICs were 4.0 μg/ml; for one strain of *Bacteroides vulgatus* the trovafloxacin MIC was 8 μg/ml. Ciprofloxacin did not inhibit any of the *B. fragilis* strains at ≤1 μg/ml and inhibited only 3% of other *B. fragilis* group isolates at that concentration. High trovafloxacin MICs (i.e., in the range of 1 to 8 μg/ml) were not necessarily correlated with higher than usual ciprofloxacin MICs.

The ciprofloxacin MICs for *Clostridium difficile* were in the range of 4 to 8 μg/ml, but those of trovafloxacin were in the range of 0.5 to 1 μg/ml. However, *C. difficile* is primarily of interest in relation to antimicrobial agent-induced pseudomembranous colitis. These data must be interpreted in the context of the level of drug achieved in the colon and the effect of the agent on the indigenous colonic flora. A similar pattern was seen with *Clostridium ramosum*, although a few strains were also resistant to trovafloxacin.

Interestingly, the highest trovafloxacin MICs (64 μg/ml) were seen for a few strains of *Sutterella wadsworthensis*. For 2

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TABLE 1. Activities of antimicrobial agents for various organisms

Organism and antimicrobial agent (no. of isolates)	MIC (μg/ml)[a]			% Susceptible[b]
	Range	50%	90%	
Bacteroides fragilis (91)				
Trovafloxacin	0.12–0.5	0.25	0.25	100, 100, 100
Ciprofloxacin	2–32	4	16	0, 14, 86
Chloramphenicol	2–4	4	4	100, 100, 100
Imipenem	0.06–>32	0.06	1	99, 99, 99
Metronidazole	0.25–2	0.5	1	100, 100, 100
Other *Bacteroides fragilis* group species (130)[c]				
Trovafloxacin	0.12–8	0.5	1	96, 99, 100
Ciprofloxacin	0.5–>64	16	32	3, 5, 25
Chloramphenicol	1–8	4	8	100, 100, 100
Imipenem	0.06–8	0.25	1	99, 100, 100
Metronidazole	0.12–2	1	1	100, 100, 100
Campylobacter gracilis (15)[d]				
Trovafloxacin	0.12–0.12	0.12	0.12	100, 100, 100
Ciprofloxacin	0.12–0.12	0.12	0.12	100, 100, 100
Chloramphenicol	0.25–4	0.5	2	100, 100, 100
Ciprofloxacin	0.12–0.12	0.12	0.12	100, 100, 100
Imipenem	0.12–1	0.5	1	100, 100, 100
Metronidazole	0.12–2	0.25	0.5	100, 100, 100
Other *Bacteroides* species (16)[e]				
Trovafloxacin	0.12–2	0.25	1	100, 100, 100
Ciprofloxacin	0.12–8	2	4	38, 81, 94
Chloramphenicol	1–8	2	4	100, 100, 100
Imipenem	0.06–1	0.06	0.5	100, 100, 100
Metronidazole	0.12–2	0.25	2	100, 100, 100
Sutterella wadsworthensis (21)[f]				
Trovafloxacin	0.12–64	0.25	0.5	90, 90, 95
Ciprofloxacin	0.25–64	0.5	1	90, 90, 90
Chloramphenicol	1–16	2	4	95, 100, 100
Imipenem	0.5–8	1	1	95, 100, 100
Metronidazole	0.12–128	2	64	76, 76, 81
Porphyromonas species (15)[g]				
Trovafloxacin	0.12–1	0.12	1	100, 100, 100
Ciprofloxacin	0.12–4	0.5	1	94, 94, 100
Chloramphenicol	0.12–2	1	2	100, 100, 100
Imipenem	0.06–0.06	0.06	0.06	100, 100, 100
Metronidazole	0.12–2	0.12	1	100, 100, 100
Prevotella species (49)[h]				
Trovafloxacin	0.12–4	1	1	98, 100, 100
Ciprofloxacin	0.5–16	1	4	61, 82, 90
Chloramphenicol	0.5–4	2	4	100, 100, 100
Imipenem	0.06–0.12	0.06	0.06	100, 100, 100
Metronidazole	0.12–2	0.5	1	100, 100, 100
Bilophila wadsworthia (24)				
Trovafloxacin	0.25–4	0.5	1	96, 100, 100
Ciprofloxacin	0.12–1	0.12	1	100, 100, 100
Chloramphenicol	4–8	4	4	100, 100, 100
Imipenem	0.06–0.12	0.12	0.12	100, 100, 100
Metronidazole	0.12–0.25	0.12	0.25	100, 100, 100
Fusobacterium nucleatum (28)				
Trovafloxacin	0.12–1	0.5	0.5	100, 100, 100
Ciprofloxacin	1–8	2	2	11, 96, 96
Chloramphenicol	0.25–1	1	1	100, 100, 100
Imipenem	0.06–0.06	0.06	0.06	100, 100, 100
Metronidazole	0.12–0.5	0.12	0.25	100, 100, 100
Fusobacterium mortiferum-Fusobacterium varium group (16)				
Trovafloxacin	0.5–4	1	4	81, 100, 100
Ciprofloxacin	1–16	2	8	25, 56, 88
Chloramphenicol	0.25–4	0.5	2	100, 100, 100
Imipenem	0.12–2	0.5	1	100, 100, 100
Metronidazole	0.12–0.25	0.12	0.25	100, 100, 100
Other *Fusobacterium* species (18)[i]				
Trovafloxacin	0.12–1	0.25	0.5	100, 100, 100
Ciprofloxacin	0.12–2	1	2	78, 100, 100

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TABLE 1—*Continued*

Organism and antimicrobial agent (no. of isolates)	MIC (μg/ml)[a]			% Susceptible[b]
	Range	50%	90%	
Chloramphenicol	0.12–2	1	1	100, 100, 100
Imipenem	0.06–0.5	0.06	0.5	100, 100, 100
Metronidazole	0.12–0.5	0.12	0.5	100, 100, 100
Clostridium difficile (15)[j]				
Trovafloxacin	0.5–1	1	1	100, 100, 100
Ciprofloxacin	4–8	8	8	0, 0, 13
Chloramphenicol	2–32	4	32	80, 87, 100
Imipenem	2–8	4	4	93, 100, 100
Metronidazole	0.12–0.25	0.25	0.25	100, 100, 100
Clostridium perfringens (20)				
Trovafloxacin	0.12–0.25	0.12	0.12	100, 100, 100
Ciprofloxacin	0.25–0.5	0.5	0.5	100, 100, 100
Chloramphenicol	2–4	2	4	100, 100, 100
Imipenem	0.06–0.25	0.06	0.25	100, 100, 100
Metronidazole	0.12–1	0.5	1	100, 100, 100
Clostridium ramosum (15)				
Trovafloxacin	0.12–8	0.5	0.5	93, 93, 100
Ciprofloxacin	1–>64	4	8	7, 40, 80
Chloramphenicol	2–32	4	4	93, 93, 100
Imipenem	0.12–2	0.25	0.5	100, 100, 100
Metronidazole	0.25–1	0.5	1	100, 100, 100
Other *Clostridium* species (11)[k]				
Trovafloxacin	0.12–4	0.25	0.5	91, 100, 100
Ciprofloxacin	0.12–64	2	64	45, 82, 82
Chloramphenicol	1–16	2	8	91, 100, 100
Imipenem	0.06–2	0.06	2	100, 100, 100
Metronidazole	0.12–0.5	0.25	0.5	100, 100, 100
Peptostreptococcus species (38)[l]				
Trovafloxacin	0.12–4	0.12	1	97, 100, 1000
Ciprofloxacin	0.12–16	1	8	68, 76, 82
Chloramphenicol	0.5–4	2	4	100, 100, 100
Imipenem	0.06–1	0.06	0.12	100, 100, 100
Metronidazole	0.12–2	0.25	1	100, 100, 100
Gram-positive rods (non-spore-forming) (35)[m]				
Trovafloxacin	0.12–8	0.5	4	86, 95, 100
Ciprofloxacin	0.12–16	1	16	53, 72, 78
Chloramphenicol	0.5–8	2	8	100, 100, 100
Imipenem	0.06–1	0.06	0.5	100, 100, 100
Metronidazole	0.25–>128	8	>128	54, 57, 57
Total (557)				
Trovafloxacin	0.12–64	0.25	1	96, 99, 100
Ciprofloxacin	0.12–>64	2	16	37, 52, 72
Chloramphenicol	0.12–32	4	8	99, 100, 100
Imipenem	0.06–>32	0.12	1	99, 100, 100
Metronidazole	0.12–>128	0.5	1	96, 96, 97

[a] 50% and 90%, MICs at which 50 and 90% of isolates are inhibited, respectively.

[b] Percent susceptible at three concentrations: for trovafloxacin, 2, 4, and 8 μg/ml; for ciprofloxacin, 1, 2, and 4 μg/ml; for chloramphenicol 8, 16, and 32 μg/ml; for imipenem, 4, 8, and 16 μg/ml; for metronidazole, 8, 16, and 32 μg/ml. Approved breakpoints for susceptibility are ≤4 μg/ml for imipenem and ≤8 μg/ml for chloramphenicol and metronidazole.

[c] *B. caccae*, *n* = 8; *B. distasonis*, *n* = 23; *B. merdae*, *n* = 2; *B. ovatus*, *n* = 18; *B. stercoris*, *n* = 5; *B. thetaiotaomicron*, *n* = 44; *B. uniformis*, *n* = 11; *B. vulgatus*, *n* = 19.

[d] Previously *Bacteroides gracilis*.

[e] *B. capillosis*, *n* = 1; *B. levii*, *n* = 1; *B. splanchnicus*, *n* = 4; *B. ureolyticus*, *n* = 1; *Bacteroides* species, *n* = 9.

[f] Previously atypical, bile-resistant *Bacteroides gracilis*, reclassified and published elsewhere (24).

[g] *P. asaccharaolytica*, *n* = 5; *P. endodontalis*, *n* = 4; *P. gingivalis*, *n* = 5; *Porphyromonas* species, *n* = 1.

[h] *P. bivia*, *n* = 4; *P. buccae*, *n* = 4; *P. corporis*, *n* = 2; *P. denticola*, *n* = 1; *P. disiens*, *n* = 2; *P. intermedia*, *n* = 17; *P. loescheii*, *n* = 8; *P. melaninogenica*, *n* = 4; *P. oralis*, *n* = 2; *P. oris*, *n* = 2; *Prevotella* species, *n* = 3.

[i] *F. gonidiaformans*, *n* = 2; *F. naviforme*, *n* = 2; *F. necrogenes*, *n* = 7; *Fusobacterium* species, *n* = 7.

[j] *C. difficile* is primarily of interest in relation to antimicrobial agent-induced pseudomembranous colitis. These data must be interpreted in the context of the level of drug achieved in the colon and the effect of the agent on the indigenous colonic flora.

[k] *C. cadaveris*, *n* = 1; *C. clostridioforme*, *n* = 1; *C. innocuum*, *n* = 1; *C. septicum*, *n* = 1; *C. sordelii*, *n* = 2; *C. sporogenes*, *n* = 1; *C. tertium*, *n* = 1; *Clostridium* species, *n* = 3.

[l] *P. anaerobius*, *n* = 9; *P. asaccharolyticus*, *n* = 6; *P. magnus*, *n* = 6; *P. micros*, *n* = 6; *P. prevotii*, *n* = 6; *Peptostreptococcus* species, *n* = 5.

[m] *Actinomyces israelii*, *n* = 2; *Actinomyces naeslundii*, *n* = 2; *Actinomyces odontolyticus*, *n* = 4; *Actinomyces* species, *n* = 3; *Eubacterium aerogenes*, *n* = 2; *Eubacterium alactolyticum*, *n* = 1; *Eubacterium lentum*, *n* = 3; *Eubacterium* species, *n* = 2; *Lactobacillus acidophilus*, *n* = 1; *Lactobacillus catenaformis*, *n* = 3; *Lactobacillus jensenii*, *n* = 2; *Lactobacillus minutus*, *n* = 2; *Lactobacillus* species, *n* = 2; *Propionibacterium acnes*, *n* = 6.

of 10 *S. wadsworthensis* strains, ciprofloxacin and trovafloxacin MICs were high. *S. wadsworthensis* was originally classified as an atypical *Campylobacter gracilis* strain. Interestingly, the MICs of both trovafloxacin and ciprofloxacin for *C. gracilis* strains were low. These organisms were reexamined and reclassified (23).

The other published data on trovafloxacin and anaerobic bacteria are very similar to our own. For all strains tested, the MIC of trovafloxacin at which 90% of strains are inhibited was 1.0 μg/ml; 99.6% of the strains were susceptible to trovafloxacin at ≤2.0 μg/ml (14). Trovafloxacin has excellent activity against anaerobes, and further studies are certainly warranted. Whether resistant strains will emerge during therapy is not yet known. The use of these agents in clinical trials will clarify their appropriateness for use in mixed infections involving anaerobic bacteria.

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